

## PCR amplification

- Phusion (our standard) master mix (25  $\mu\text{l}$ ):
  - 5  $\mu\text{l}$  5 x HF-buffer
  - 0.5  $\mu\text{l}$  10mM dNTPs
  - Up to 20 ng \*  $\mu\text{l}^{-1}$  template
  - 0.25  $\mu\text{l}$  polymerase
  - 0.25-0.5  $\mu\text{l}$  primer 10  $\mu\text{M}$  (each one)
  - 0.75  $\mu\text{l}$  DMSO (optional)
  - Fill up to 25  $\mu\text{l}$  with  $\text{H}_2\text{O}$
- Q5 master mix (25  $\mu\text{l}$ ):
  - 5  $\mu\text{l}$  5 x buffer
  - 0.5  $\mu\text{l}$  10 mM dNTPs
  - Up to 50 ng \*  $\mu\text{l}^{-1}$  template
  - 0.1  $\mu\text{l}$  polymerase
  - 0.5-1  $\mu\text{l}$  primer 10  $\mu\text{M}$  (each one)
  - Fill up to 25  $\mu\text{l}$  with  $\text{H}_2\text{O}$
- KOD master mix (50  $\mu\text{l}$ ):
  - 5  $\mu\text{l}$  10 x buffer for KOD Hot Start DNA Polymerase
  - 3  $\mu\text{l}$  25 mM  $\text{MgSO}_4$
  - 1  $\mu\text{l}$  10mM dNTPs
  - 1  $\mu\text{l}$  polymerase
  - 1.5  $\mu\text{l}$  primer 10  $\mu\text{M}$  (each one)
  - 10 ng \*  $\mu\text{l}^{-1}$  template
  - Fill up to 50  $\mu\text{l}$  with  $\text{H}_2\text{O}$

